Lipoprotein(a) levels, apo(a) isoform size, and coronary heart disease risk in the Framingham Offspring Study

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Lipoprotein(a) (Lp(a)) was first described by Berg in 1965 (1). Lp(a) is a lipoprotein similar in structure to LDL, but differs from LDL in having apo(a), a large glycoprotein, attached to apoB-100 by a disulfide bond (2). Apo(a) shares strong homology with several regions of plasminogen (3), including the protease domain, the kringle 5 domain, and 10 types of the kringle 4 domain. It has been shown that apo(a) is highly polymorphic in size due to different numbers of the kringle 4 type 2 (kringle-42) domain, ranging from a minimum of 3 to greater than 40 (4). Plasma levels of Lp(a) are highly heritable (2, 5) and are inversely correlated with the apo(a) isoform size, with subjects carrying small isoforms having high plasma Lp(a) levels (5). Metabolic studies have shown that the inverse association between plasma Lp(a) levels and apo(a) isoform size is due to differences in the hepatic secretion of apo(a), with subjects carrying small apo(a) isoforms having increased apo(a) secretion (6, 7). Boerwinkle et al. (8) indicated that 69% of the variability in plasma Lp(a) levels is accounted for by the number of apo(a) kringle-42 repeats, and an additional 21% of the variation is explained by other sequences within the gene coding for apo(a). Recently, the rs3798220 single-nucleotide polymorphism (SNP) in the apo(a) locus was found to be a strong predictor of Lp(a) levels and coronary heart disease (CHD) risk (9). Plasma Lp(a) levels are associated with CHD risk (10, 11). The mechanisms by which Lp(a) increases CHD risk are not well defined, but may include both a prothrombotic effect due to the similarity of apo(a) to the fibrinolytic pro-enzyme plasminogen (12, 13), and an atherogenic effect mediated by the preferential binding of oxidized phospholipids by Lp(a) as well as Lp(a) deposition in the arterial wall (14).

Abstract The aim of this study was to assess the independent contributions of plasma levels of lipoprotein(a) (Lp(a)), Lp(a) cholesterol, and of apo(a) isoform size to prospective coronary heart disease (CHD) risk. Plasma Lp(a) and Lp(a) cholesterol levels, and apo(a) isoform size were measured at examination cycle 5 in subjects participating in the Framingham Offspring Study who were free of CHD. After a mean follow-up of 12.3 years, 98 men and 47 women developed new CHD events. In multivariate analysis, the hazard ratio of CHD was approximately two-fold greater in men in the upper tertile of plasma Lp(a) levels, relative to those in the bottom tertile (P < 0.002). The apo(a) isoform size contributed only modestly to the association between Lp(a) and CHD and was not an independent predictor of CHD. In multivariate analysis, Lp(a) cholesterol was not significantly associated with CHD risk in men. In women, no association between Lp(a) and CHD risk was observed. Elevated plasma Lp(a) levels are a significant and independent predictor of CHD risk in men. The assessment of apo(a) isoform size in this cohort does not add significant information about CHD risk. In addition, the cholesterol content in Lp(a) is not a significant predictor of CHD risk.—Lamon-Fava, S., S. M. Marcovina, J. J. Albers, H. Kennedy, C. DeLuca, C. C. White, L. A. Cupples, J. R. McNamara, L. J. Seman, V. Bongard, and E. J. Schaefer. Lipoprotein(a) levels, apo(a) isoform size, and coronary heart disease risk in the Framingham Offspring Study. J. Lipid Res. 2011. 52: 1181–1187.

Supplementary key words apolipoprotein • cholesterol • epidemiology • triglycerides
The goal of this study was to assess prospectively the risk of CHD associated with elevated plasma Lp(a) levels and the contribution of apo(a) isoform size to this association in the Framingham Offspring Study (FOS). In addition, we have compared the CHD risk prediction associated with elevated Lp(a) levels as assessed by two different immunoassays and by the measurement of Lp(a) cholesterol levels.

METHODS

Subjects

The FOS is a longitudinal population-based study which started in 1975 and enrolled the children of the participants in the original Framingham Heart Study cohort, and their spouses (15). All participants are Caucasian. During the 5th examination cycle (1991–1995), subjects underwent a medical history, physical examination, electrocardiogram (ECG) evaluation, and a blood draw for the assessment of plasma lipid and lipoprotein levels. At cycle 5, there were 1,328 men and 1,562 women who were free of CHD and had their plasma Lp(a) levels measured. These subjects were followed until completion of cycle 8, with a mean follow-up of 12.3 years, for the occurrence of new CHD events. All suspected CHD events were assessed by a panel of three physicians who evaluated the available evidence. CHD was defined as: myocardial infarction diagnosed by ECG, myocardial infarction diagnosed by enzymatic elevations and history, sudden CHD death, and non-sudden CHD death.

Lp(a) assessment

Blood was drawn after a 12 h overnight fast into 0.1% EDTA tubes. Plasma was separated by centrifugation at 2,500 rpm for 30 min at 4°C and immediately frozen and stored at −80°C. Plasma levels of total cholesterol (TC), triglycerides (TGs), and HDL-cholesterol (HDL-C) were measured by automated enzymatic methods in an Abbott Diagnostics ABA-200 analyzer using Abbott A-Gent enzymatic reagents. LDL-cholesterol (LDL-C) levels were calculated using the Friedewald formula when TG levels were <400 mg/dl (16).

Lp(a) protein levels were measured using an ELISA. This assay utilizes as capturing antibody a monoclonal antibody against apo(a) that does not recognize the kringle-4-2 domain and is independent of apo(a) isoform size (17). Lp(a) protein levels are expressed in nmol/l. The Lp(a) standard used for this assay was a lyophilized serum having kringles 18 and 25 and an assigned value of 123 nmol/l using World Health Organization/International Federation of Clinical Chemistry and Laboratory Medicine reference material (18). Inter- and intra-assay coefficients of variation were 3.2% and 3.5%, respectively. In some individuals, only one apo(a) isoform is expressed. However, in most individuals, apo(a) is expressed by both alleles, with one isoform more prevalent than the other. Apo(a) isoforms were measured with methodology previously described (4), in which the size of apo(a) isoforms is visualized on agarose gel electrophoresis. The apo(a) isoform size thus visualized is directly proportional to the number of kringle-4-2 repeats (4). The predominant apo(a) isoform size was used in statistical analysis. These measurements were carried out at the Northwest Lipid Metabolism and Diabetes Research Laboratories (NWRL), University of Washington, Seattle, WA.

Lp(a) levels were also measured with two other assays in plasma obtained at examination cycle 5: an immunoturbidimetric assay from Wako Chemicals USA (Richmond, VA), which utilizes an antibody that does not cross-react with plasminogen or apoB (inter-and intra-assay coefficients of variation (CVs) <3%), and a lectin-based assay (Genzyme Diagnostics; Framingham, MA), which utilizes lectin to capture Lp(a) and then measures the cholesterol content in the particle (inter-and intra-assay CV: 3.8% and 3.2%, respectively) (19). Commercially available standards were used (Wako Chemicals USA). Measurements with these two assays were carried out at the Lipid Metabolism Laboratory, Tufts University, Boston, MA.

The genotyping for the rs3798220 SNP, located in the apo(a) locus, was performed according to established methodology (20).

Statistical analysis

Variables were assessed for normal distribution, and a log-transformation was applied to skewed variables before analysis. Means and standard deviations are reported for normally distributed variables, and median and inter-quartile ranges are reported for skewed variables. Analyses were stratified by sex. Differences in baseline characteristics for subjects who developed new CHD events during follow-up (CHD cases) versus subjects who did not develop CHD (controls) were compared using Student’s t-tests for continuous variables and χ² tests for dichotomous variables. Cox proportional hazards models were used to estimate the hazard ratio (HR) of CHD associated with high levels of Lp(a). In these models, subjects in the upper tertile of Lp(a) levels (or lower tertile of apo(a) isoform size) were compared with subjects in the lower tertile of Lp(a) (or upper tertile of apo(a) isoform size). Models were adjusted for possible confounding risk factors for CHD, including age, body mass index (BMI), smoking, hypertension, diabetes, TC, TG, HDL-C, and use of lipid-lowering medications.

RESULTS

Plasma Lp(a) concentrations were measured with three different assays in FOS participants at examination cycle 5. The percentile distribution of Lp(a) concentrations measured with the different methods is illustrated in Table 1. The Lp(a) protein values obtained by the NWRL ELISA were highly correlated to the Lp(a) concentrations obtained by the Wako immunoturbidimetric assay (R = 0.930, P < 0.0001). However, after conversion of the Lp(a) Wako measurements from mg/dl to nmol/l using a factor based on the predominant allele size, this assay showed a strong apo(a) isoform size-dependent bias (Fig. 1), consistent with the concept that the Wako assay overestimates the Lp(a) values in samples with large apo(a) isoforms and underestimates the Lp(a) values in samples with small apo(a) isoforms. A bias greater than 10% was detected in 73% of the population. The cholesterol content of Lp(a), measured using the lectin-affinity chromatography method, was correlated with both the NWLR Lp(a) protein and the Wako Lp(a) concentrations (R = 0.765, P < 0.0001 and R = 0.789, P < 0.0001, respectively). However, a comparison of the Lp(a) cholesterol values with the other Lp(a) measurements, based on percentiles shown in Table 1, suggested that the Lp(a) cholesterol method may overestimate the amount of Lp(a) cholesterol in samples with low Lp(a) values. The predominant apo(a) isoform size was significantly and inversely related to plasma levels of Lp(a) as assessed with the different assays (NWRL method,
Wake method, a greater number of CHD cases, relative to controls, were observed in men in the upper tertile of plasma Lp(a) levels as compared with men in the lower tertile (Table 3). This was not observed in women (Table 3).

In multivariate Cox proportional hazards models, the HR of incident CHD was greater than 2-fold in men in the upper tertile of Lp(a) levels than in men in the lower tertile (Table 4), both with the NWRL and the Wako Lp(a) measurements. The risk of CHD associated with high Lp(a) levels was not modified after adjusting for several risk factors for CHD, including age, BMI, smoking, hypertension, diabetes, and plasma levels of TC, log-TG, and HDL-C (Table 4, model 3), or further adjustment for use of cholesterol-lowering medications and niacin [a medication known to affect plasma Lp(a) levels] (Table 4, model 4). Apo(a) isoform size is known to be a significant determinant of plasma Lp(a) levels, and after further adjustment for the predominant isoform size, the association between high Lp(a) levels and CHD was still significant (Table 4, model 5). In separate multivariate models, the HR for incident CHD was significantly higher in men in the lower tertile of apo(a) isoform size than in those in the higher tertile (Table 4). However, the strength of this association was lower than observed for Lp(a) levels, and the significance was abolished after adjustment for plasma Lp(a) levels measured with either the NWRL or Wako method, a greater number of CHD cases, relative to controls, were observed in men in the upper tertile of plasma Lp(a) levels as compared with men in the lower tertile (Table 3). This was not observed in women (Table 3).

When subjects were divided according to tertiles of plasma Lp(a) levels, as assessed by either the NWRL or Wake method, a greater number of CHD cases, relative to controls, were observed in men in the upper tertile of plasma Lp(a) levels as compared with men in the lower tertile (Table 3). This was not observed in women (Table 3).

Men who developed new CHD events during the 12.3 years follow-up were significantly older and had a higher prevalence of CHD risk factors than men who did not develop CHD (Table 2). Median plasma Lp(a) levels were approximately two-fold higher in male CHD cases than in male control subjects for both immuno-based Lp(a) methods (NWRL, \( P = 0.003 \); and Wako, \( P = 0.01 \)) (Table 2). The difference in median plasma levels of Lp(a) cholesterol between men with incident CHD and controls was marginally significant (\( P = 0.045 \)). The average predominant apo(a) isoform size was significantly smaller in male CHD cases than in controls (\( P = 0.025 \)) (Table 2). During follow-up, only 3.1% of women developed CHD, as opposed to 7.9% of men. Women with CHD, similar to men, were significantly older and had a higher prevalence of CHD risk factors than women without CHD (Table 2). The difference in plasma lipid levels and in CHD risk factors between cases and controls was more marked in women than in men. In contrast to men, median plasma Lp(a) protein, Lp(a), or Lp(a) cholesterol levels were not significantly different between women CHD cases and controls, and the average predominant apo(a) isoform was marginally larger in cases than in controls (\( P = 0.044 \)).

When subjects were divided according to tertiles of plasma Lp(a) levels, as assessed by either the NWRL or
by immunoassay or electrophoresis, were significantly associated with CHD risk (21–23). Recently, there has been a resurgence of interest in Lp(a), with studies conducted in large numbers of subjects indicating an independent association of Lp(a) levels with CHD risk. The Emerging Risk Factors Collaboration group has reported a significant and curvilinear association between plasma Lp(a) levels and CHD, defined as first MI or CHD death, in 126,634 participants in 36 different prospective studies (11). In this study, there was no association between plasma Lp(a) levels and nonvascular death (11). In addition, a meta-analysis of 31 published prospective studies in which baseline Lp(a) levels were measured with somewhat different methodologies indicated an odds ratio of 1.45 (CI = 1.32–1.58) for CHD in subjects in the upper tertile of Lp(a) levels, relative to subjects in the lower tertile (10).

The allele frequency of the rs3798220 SNP was 1.56 in men and 1.55 in women. This SNP was associated with plasma Lp(a) levels both in men and women (P < 0.0001). However, in the fully adjusted multivariate Cox proportional hazards models, this SNP was not significantly associated with CHD risk in men (HR = 1.29, CI = 0.39–4.21, P = 0.66) or women (HR = 1.53, CI = 0.36–6.54, P = 0.56).

**DISCUSSION**

Elevated levels of Lp(a) have long been known to be associated with premature CHD. In cross-sectional analysis of the Lipid Research Clinics Coronary Primary Prevention Trial and the Framingham Heart Study, we had previously shown that elevated levels of Lp(a), whether assessed by immunoassay or electrophoresis, were significantly associated with CHD risk (21–23). Recently, there has been a resurgence of interest in Lp(a), with studies conducted in large numbers of subjects indicating an independent association of Lp(a) levels with CHD risk. The Emerging Risk Factors Collaboration group has reported a significant and curvilinear association between plasma Lp(a) levels and CHD, defined as first MI or CHD death, in 126,634 participants in 36 different prospective studies (11). In this study, there was no association between plasma Lp(a) levels and nonvascular death (11). In addition, a meta-analysis of 31 published prospective studies in which baseline Lp(a) levels were measured with somewhat different methodologies indicated an odds ratio of 1.45 (CI = 1.32–1.58) for CHD in subjects in the upper tertile of Lp(a) levels, relative to subjects in the lower tertile (10). Our re-

**TABLE 2.** Baseline characteristics, plasma lipid and Lp(a) levels, and predominant apo(a) isoform size in subjects who developed CHD during a mean follow-up of 12.3 years, and in subjects free of CHD

<table>
<thead>
<tr>
<th>Variable</th>
<th>Controls</th>
<th>CHD cases</th>
<th>P</th>
<th>Controls</th>
<th>CHD cases</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n = 1,230</td>
<td>n = 98</td>
<td></td>
<td>n = 1,505</td>
<td>n = 47</td>
<td></td>
</tr>
<tr>
<td>Age, y</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>BMI, kg/m²</td>
<td>28.17 ± 4.25</td>
<td>28.95 ± 3.49</td>
<td>0.046</td>
<td>26.74 ± 5.41</td>
<td>28.27 ± 5.66</td>
<td>0.057</td>
</tr>
<tr>
<td>TC, mg/dl</td>
<td>201 ± 33</td>
<td>207 ± 39</td>
<td>0.117</td>
<td>208 ± 37</td>
<td>232 ± 37</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>TG, mg/dl</td>
<td>128 [88, 192]</td>
<td>149 [104, 207]</td>
<td>0.011*</td>
<td>112 [81, 162]</td>
<td>172 [106, 223]</td>
<td>&lt;0.0001*</td>
</tr>
<tr>
<td>LDL-C, mg/dl</td>
<td>121 ± 30</td>
<td>124 ± 34</td>
<td>0.457</td>
<td>118 ± 33</td>
<td>141 ± 31</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>HDL-C, mg/dl</td>
<td>43 ± 11</td>
<td>41 ± 10</td>
<td>0.646</td>
<td>56 ± 16</td>
<td>47 ± 12</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Lp(a) protein, nmol/L</td>
<td>18.1 [4.4, 65]</td>
<td>36.3 [10.6, 124.4]</td>
<td>0.005*</td>
<td>20.6 [5.6, 71.3]</td>
<td>18.8 [4.4, 53.1]</td>
<td>0.353*</td>
</tr>
<tr>
<td>Lp(a), mg/dl (Wako)</td>
<td>10.8 [4.9, 30]</td>
<td>20.6 [7.6, 38]</td>
<td>0.010*</td>
<td>13 [5.9, 34]</td>
<td>12 [5.5, 56]</td>
<td>0.71*</td>
</tr>
<tr>
<td>Lp(a)-C, mg/dl (Genzyme)</td>
<td>4.9 [2.6, 9.5]</td>
<td>5.9 [3.2, 11.6]</td>
<td>0.045*</td>
<td>4.7 [2.5, 9.3]</td>
<td>5.5 [2.6, 12.2]</td>
<td>0.49*</td>
</tr>
<tr>
<td>Predominant isoform</td>
<td>24.9 ± 5.5</td>
<td>22.7 ± 5.5</td>
<td>0.025</td>
<td>23.9 ± 5.6</td>
<td>25.6 ± 6.2</td>
<td>0.044</td>
</tr>
<tr>
<td>Systolic blood pressure, mm Hg</td>
<td>129 ± 17</td>
<td>134 ± 18</td>
<td>0.006</td>
<td>124 ± 20</td>
<td>138 ± 21</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Diastolic blood pressure, mm Hg</td>
<td>77 ± 10</td>
<td>77 ± 10</td>
<td>0.689</td>
<td>73 ± 10</td>
<td>77 ± 9</td>
<td>0.005</td>
</tr>
<tr>
<td>Hypertension, %</td>
<td>34</td>
<td>55</td>
<td>&lt;0.0001</td>
<td>29</td>
<td>62</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Smoking, %</td>
<td>19</td>
<td>24</td>
<td>0.307</td>
<td>19</td>
<td>37</td>
<td>0.003</td>
</tr>
<tr>
<td>Diabetes, %</td>
<td>8</td>
<td>18</td>
<td>0.001</td>
<td>5</td>
<td>17</td>
<td>0.0002</td>
</tr>
<tr>
<td>Lipid-lowering medications, %</td>
<td>7</td>
<td>19</td>
<td>&lt;0.0001</td>
<td>5</td>
<td>13</td>
<td>0.030</td>
</tr>
<tr>
<td>Postmenopausal, %</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hormone therapy, %</td>
<td>17</td>
<td></td>
<td>0.436</td>
<td>17</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Data shown as mean ± SD, with the exception of TG and Lp(a), where values are shown as median [IQR]. HDL-C, HDL-cholesterol; LDL-C, LDL cholesterol.

* P-value after variable was log-transformed.

**TABLE 3.** Number of new CHD cases by tertiles of baseline plasma Lp(a) values

<table>
<thead>
<tr>
<th>Lp(a) tertiles (NWRL)</th>
<th>Lp(a) tertiles (Wako)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1 (0.6–8.1 nmol/L)</td>
</tr>
<tr>
<td></td>
<td>2 (8.8–38.8 nmol/L)</td>
</tr>
<tr>
<td></td>
<td>3 (39.4–469.4 nmol/L)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Men</th>
<th>Women</th>
</tr>
</thead>
<tbody>
<tr>
<td>Controls, n</td>
<td>Controls, n</td>
</tr>
<tr>
<td>420</td>
<td>503</td>
</tr>
<tr>
<td>CHD cases, n</td>
<td>CHD cases, n</td>
</tr>
<tr>
<td>21</td>
<td>17</td>
</tr>
<tr>
<td>CHD cases, %</td>
<td>CHD cases, %</td>
</tr>
<tr>
<td>4.8</td>
<td>3.3</td>
</tr>
</tbody>
</table>

DISCUSSION

Elevated levels of Lp(a) have long been known to be associated with premature CHD. In cross-sectional analysis of the Lipid Research Clinics Coronary Primary Prevention Trial and the Framingham Heart Study, we had previously shown that elevated levels of Lp(a), whether assessed by immunoassay or electrophoresis, were significantly associated with CHD risk (21–23). Recently, there has been a resurgence of interest in Lp(a), with studies conducted in large numbers of subjects indicating an independent association of Lp(a) levels with CHD risk. The Emerging Risk Factors Collaboration group has reported a significant and curvilinear association between plasma Lp(a) levels and CHD, defined as first MI or CHD death, in 126,634 participants in 36 different prospective studies (11). In this study, there was no association between plasma Lp(a) levels and nonvascular death (11). In addition, a meta-analysis of 31 published prospective studies in which baseline Lp(a) levels were measured with somewhat different methodologies indicated an odds ratio of 1.45 (CI = 1.32–1.58) for CHD in subjects in the upper tertile of Lp(a) levels, relative to subjects in the lower tertile (10). Our re-
after further adjustment for Lp(a) levels, the association
sus the highest quartile. In one of the three populations, a
small case-control study, Kraft et al. (26) have reported
to isoform size from that of plasma levels of Lp(a). In a
studies have attempted to separate the prediction relative
to isoform size in risk prediction in the Framingham cohort.
plasma Lp(a) levels are more informative than is apo(a)
comparable results. This fi nding emphasizes the fact that
size. Similar models using both isoform sizes provided
Lp(a) levels abolished the predictive value of the isoform
was the primary variable, models adjusting for plasma
form size was entered into the hazard prediction model.
In addition, when the predominant apo(a) isoform size
was the primary variable, models adjusting for plasma
Lp(a) levels remained statistically significant after adjustment for other lipoprotein levels and other established risk factors for CHD, supporting the independent role of Lp(a) in CHD develop-
ment. In men, the association between plasma Lp(a) concentra-
tions and CHD remained significant after iso-
form size was entered into the hazard prediction model. In
addition, when the predominant apo(a) isoform size
was the primary variable, models adjusting for plasma
Lp(a) levels abolished the predictive value of the isoform
size. Similar models using both isoform sizes provided
comparable results. This finding emphasizes the fact that
plasma Lp(a) levels are more informative than is apo(a)
isoform size in risk prediction in the Framingham cohort.

Previous studies have examined the role of apo(a) iso-
form size on CHD prediction (24–30), and some of these
studies have attempted to separate the prediction relative
to isoform size from that of plasma levels of Lp(a). In a
small case-control study, Kraft et al. (26) have reported
that both Lp(a) levels and isoform size were associated
with CHD, but when both variables were entered in the
multivariate model, the strength of the association was
higher for apo(a) isoform size. Similarly, Rifai et al. (29)
have found both Lp(a) levels and apo(a) isoform size to
be significant predictors of CHD in a small case-control
study, and when both variables were entered in the same
multivariate model, only apo(a) isoform size remained sig-
nificantly associated with CHD. In three separate Danish
populations, Kamstrup et al. (28) have reported an in-
creased risk of CHD (HR = 1.3 to 1.4) in subjects in the
lowest quartile of number of apo(a) kringle-4 repeats ver-
sus the highest quartile. In one of the three populations,
after further adjustment for Lp(a) levels, the association
between the number of kringle-4 repeats was attenuated
but remained significant (28). It has been suggested that
the stronger association between apo(a) isoform and CHD
could be explained by a greater atherogenicity of Lp(a)
particles with lower number of repeats (28, 29). However,
in two Asian Indian populations, Lp(a) levels were associ-
ated with CHD risk independently of apo(a) isoform size
(30). In our prospective study, the multivariate analysis in-
cluding Lp(a) and apo(a) isoform size in the same model
clearly indicates that the majority of the increased risk of
CHD by Lp(a) in men is attributable to the concentration of
the lipoprotein particle, and that isoform size contrib-
utes to a smaller degree. The contribution of isoform size
to risk prediction could be explained by isoform size being
one, but not the only, determinant of plasma Lp(a) lev-
els, and further supports the concept that Lp(a) con-
centrations are directly associated with CHD risk.

Our comparison of different Lp(a) assays indicates that
the two immuno-based assays had similar CHD prediction
value. However, it should be pointed out that the Lp(a)
concentrations as measured by the Wako assay were over-
estimated up to 130% in subjects with very large predomi-
nant apo(a) isoform sizes, and underestimated up to 37%
in subjects with small isoform sizes. On the other hand, the
cholesterol content of Lp(a) was not significantly associated

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**TABLE 4. Hazard ratios for incident CHD in FOS men by baseline Lp(a) levels (upper tertile vs. lower tertile) or apo(a) isoform size (lower tertile vs. upper tertile)**

<table>
<thead>
<tr>
<th>Model</th>
<th>HR (Lp(a) (NWRL))</th>
<th>CI</th>
<th>P</th>
<th>HR (Lp(a) (Wako))</th>
<th>CI</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2.23</td>
<td>1.35–4.74</td>
<td>0.005</td>
<td>2.36</td>
<td>1.36–4.09</td>
<td>0.003</td>
</tr>
<tr>
<td>2</td>
<td>2.48</td>
<td>1.46–4.21</td>
<td>0.0008</td>
<td>2.69</td>
<td>1.51–4.78</td>
<td>0.0008</td>
</tr>
<tr>
<td>3</td>
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<td>1.50–4.40</td>
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<td>2.69</td>
<td>1.51–4.81</td>
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<td>4</td>
<td>2.36</td>
<td>1.37–4.07</td>
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<td>2.52</td>
<td>1.40–4.53</td>
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<tr>
<td>5</td>
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<td>1.20–4.99</td>
<td>0.013</td>
<td>2.74</td>
<td>1.35–5.58</td>
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</tr>
<tr>
<td></td>
<td>Predominant apo(a) isoform</td>
<td></td>
<td></td>
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<tr>
<td>1</td>
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<td>1.04–2.77</td>
<td>0.03</td>
<td>1.70</td>
<td>1.04–2.77</td>
<td>0.03</td>
</tr>
<tr>
<td>2</td>
<td>1.76</td>
<td>1.07–2.88</td>
<td>0.03</td>
<td>1.76</td>
<td>1.07–2.88</td>
<td>0.03</td>
</tr>
<tr>
<td>3</td>
<td>1.78</td>
<td>1.09–2.93</td>
<td>0.03</td>
<td>1.78</td>
<td>1.09–2.93</td>
<td>0.03</td>
</tr>
<tr>
<td>4</td>
<td>1.68</td>
<td>1.02–2.77</td>
<td>0.04</td>
<td>1.68</td>
<td>1.02–2.77</td>
<td>0.04</td>
</tr>
<tr>
<td>6</td>
<td>1.15</td>
<td>0.59–2.25</td>
<td>0.67</td>
<td>1.08</td>
<td>0.54–2.12</td>
<td>0.83</td>
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</table>

Model 1 covariate: age; Model 2 covariates: age, BMI, smoking, hypertension, and diabetes; Model 3 covariates: age, BMI, smoking, hypertension, diabetes, TC, log-TG, and HDL-C; Model 4 covariates: age, BMI, smoking, hypertension, diabetes, TC, log-TG, HDL-C, and use of cholesterol-lowering medications and/or niacin; Model 5 covariates as in Model 4, plus predominant apo(a) isoform size; Model 6 covariates as in Model 4, plus Lp(a) levels.
with CHD in multivariate analysis. This may suggest that either the cholesterol content of the particle, which is influenced by the metabolism of the particle, similar to the cholesterol content in LDL, does not entirely account for the atherogenicity of Lp(a), or that the methodology for Lp(a) cholesterol assessment is not accurate. Although our and other works confirm the causative link between elevated Lp(a) and CHD, the mechanism responsible for the atherogenicity of Lp(a) is still not clear. Lp(a) has been shown to serve as a scavenger of oxidized phospholipids (14, 33), and this functional property may be responsible for proatherogenic and pro-inflammatory effects of Lp(a). Also, due to the high degree of homology between apo(a) and plasminogen (3), it has been hypothesized that Lp(a) interferes with the fibrinolytic process and thus may be prothrombotic (34). Both of these putative functions of Lp(a) are attributed to the apo(a) component.

In women, we did not find a significant association between CHD and Lp(a) levels. This is in contrast to other large prospective and case-control studies indicating a similar association between high Lp(a) levels and CHD risk in men and women (9, 11). Our power calculation analysis indicated that both a higher event rate and a greater number of participants would have been needed to detect a significant association between Lp(a) levels and CHD risk in women in the FOS.

Our study supports a significant and independent role of Lp(a) on cardiovascular disease in men.

REFERENCES


